

WHAT IS CLAIMED IS:

1. A primer selected from the group of:

5' - TGC TTA ATC AGT GAG GCA CC - 3' (SEQ ID NO:1);

5' - AGA TCA GTT GGG TGC ACG AG - 3' (SEQ ID NO:2);

5' - CTT GGT CTG ACA GTT ACC - 3' (SEQ ID NO:3);

5' - TGT CGC CCT TAT TCC - 3' (SEQ ID NO:4); and

5' - TCG GGG AAA TGT GCG - 3' (SEQ ID NO:5).

2. A primer selected from the group of:

5' - ATC GTC CAC CAT CCA CTG CA - 3' (SEQ ID NO:6);

5' - GGG AAA CGG AAC TGA ATG AG - 3' (SEQ ID NO:7);

5' - TAG TGG ATC TTT CGC TCC AG - 3' (SEQ ID NO:8);

5' - GCT CTG CTT TGT TAT TC - 3' (SEQ ID NO:9);

5' - CAC TCA AGG ATG TAT TGT G - 3' (SEQ ID NO:10); and

5' - TTA GCG TTG CCA GTG CTC G - 3' (SEQ ID NO:11).

3. A primer selected from the group of:

5' - GGA ACA GAC TGG GCT TTC ATC - 3' (SEQ ID NO:12);

5' - GGA CAT CCC CTT GAC - 3' (SEQ ID NO:13);

5' - GTG GAT TCA CTT CTG CCA CG - 3' (SEQ ID NO:14);

5' - CTT CTG GCA TGC CCT ATG AG - 3' (SEQ ID NO:15);

5' - CAT GAC CCA GTT CGC CAT ATC CTG - 3' (SEQ ID NO:16);

5' - ATT CGT ATG CTG GAT CTC GCC ACC - 3' (SEQ ID NO:17);

5' - CGA ACG AAT CAT TCA GCA CCG - 3' (SEQ ID NO:44); and

5' - CGG CAA TGT TTT ACT GTA GCG CC - 3' (SEQ ID NO:45).

5 4. A primer selected from the group of:

5' - CTG GCA ACC ACA ATG GAC TCC G - 3' (SEQ ID NO:18); and

5' - GCC AGT TCA GCA TCT CCC AGC C - 3' (SEQ ID NO:19).

5. A primer selected from the group of:

10 5' - CGT GAC CAA CAA CGC CCA GC - 3' (SEQ ID NO:20); and

5' - CCA GAT AGC GAA TCA GAT CGC - 3' (SEQ ID NO:21).

6. A primer selected from the group of:

15 5' - CCA GCC GAT GCT CAA GGA G - 3' (SEQ ID NO:22); and

5' - CAC GAA CGC CAC ATA GGC G - 3' (SEQ ID NO:23).

7. A primer selected from the group of:

5' - GGC ATT GGG ATA GTT GCG GTT G - 3' (SEQ ID NO:24); and

5' - TTA CTA CAA GGT CGG CGA CAT GAC C - 3' (SEQ ID NO:25).

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8. A primer selected from the group of:

5' - GGA TCA CAC TAT TAC ATC TCG C - 3' (SEQ ID NO:26); and

5' - CGT ATG GTT GAG TTT GAG TGG C - 3' (SEQ ID NO:27).

9. A primer selected from the group of:

5' - GCG ACC TGG TTA ACT ACA ATC CC - 3' (SEQ ID NO:28); and

5' - CGG TAG TAT TGC CC TTA AGC C - 3' (SEQ ID NO:29).

5 10. A primer selected from the group of:

5' - CGG AAA AGC ACG TCG ATG GG - 3' (SEQ ID NO:30); and

5' - GCG ATA TCG TTG GTG GTG CC - 3' (SEQ ID NO:31).

11. A primer selected from the group of:

10 5' - CTC GAT GAT GCG TGC TTC GC - 3' (SEQ ID NO:32); and

5' - GCG ACT GTG ATG TAT AAA CG - 3' (SEQ ID NO:33).

12. A primer selected from the group of:

5' - CGT CGC TCA CCA TAT CTC CC - 3' (SEQ ID NO:34); and

15 5' - CCT CTC GTG CTT TAG ACC CG - 3' (SEQ ID NO:35).

13. A primer selected from the group of:

5' - CGC TGG GAA ACC TAT TCG G - 3' (SEQ ID NO:36); and

5' - CTG CCA TCC AGT TTC TTC GGG - 3' (SEQ ID NO:37).

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14. A primer selected from the group of:

5' - GGT GGC ATT GAC AAA TTC TGG - 3' (SEQ ID NO:38); and

5' - CCC ACC ATG CGA CAC CAG - 3' (SEQ ID NO:39).

15. A primer selected from the group of:

5' - TGT GCA ACG CAA ATG GCA C - 3' (SEQ ID NO:40); and

5' - CGA CCC CAA GTT TCC TGT AAG TG - 3' (SEQ ID NO:41).

5 16. A primer selected from the group of:

5' - AGG CAC GAT AGT TGT GGC AGA C - 3' (SEQ ID NO:42); and

5' - CAC TCA ACC CAT CCT ACC CAC C - 3' (SEQ ID NO:43).

10 17. A method for identifying a beta-lactamase in a clinical sample, the method comprising:

providing a pair of oligonucleotide primers, wherein one primer of the pair is complementary to at least a portion of the beta-lactamase nucleic acid in the sense strand and the other primer of each pair is complementary to at least a portion of the beta-lactamase nucleic acid in the antisense strand;

15 annealing the primers to the beta-lactamase nucleic acid;

simultaneously extending the annealed primers from a 3' terminus of each primer to synthesize an extension product that is complementary to the nucleic acid strands annealed to each primer wherein each extension product after separation from the beta-lactamase nucleic acid serves as a template for the synthesis of an extension product for the other primer of each pair;

20 separating the amplified products; and

analyzing the separated amplified products for a region characteristic of the beta-lactamase.

25 18. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the TEM family of beta-lactamase enzymes.

19. The method of claim 18 wherein the primers are selected from the group of:

5' - TGC TTA ATC AGT GAG GCA CC - 3' (SEQ ID NO:1);

5' - AGA TCA GTT GGG TGC ACG AG - 3' (SEQ ID NO:2);

5' - CTT GGT CTG ACA GTT ACC - 3' (SEQ ID NO:3);

5' - TGT CGC CCT TAT TCC - 3' (SEQ ID NO:4); and

5' - TCG GGG AAA TGT GCG - 3' (SEQ ID NO:5);

20. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the SHV family of beta-lactamase enzymes.

21. The method of claim 20 wherein the primers are selected from the group of:

5' - ATC GTC CAC CAT CCA CTG CA - 3' (SEQ ID NO:6);

5' - GGG AAA CGG AAC TGA ATG AG - 3' (SEQ ID NO:7);

5' - TAG TGG ATC TTT CGC TCC AG - 3' (SEQ ID NO:8);

5' - GCT CTG CTT TGT TAT TC - 3' (SEQ ID NO:9);

5' - CAC TCA AGG ATG TAT TGT G - 3' (SEQ ID NO:10); and

5' - TTA GCG TTG CCA GTG CTC G - 3' (SEQ ID NO:11).

22. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the AmpC beta-lactamase enzyme found in *Enterobacter cloacae*.

23. The method of claim 22 wherein the primers are selected from the group of:

5' - GGA ACA GAC TGG GCT TTC ATC - 3' (SEQ ID NO:12);

5' - GGA CAT CCC CTT GAC - 3' (SEQ ID NO:13);

5' - GTG GAT TCA CTT CTG CCA CG - 3' (SEQ ID NO:14);  
5' - CTT CTG GCA TGC CCT ATG AG - 3' (SEQ ID NO:15);  
5' - CAT GAC CCA GTT CGC CAT ATC CTG - 3' (SEQ ID NO:16); and  
5' - ATT CGT ATG CTG GAT CTC GCC ACC - 3' (SEQ ID NO:17).  
5' - CGA ACG AAT CAT TCA GCA CCG - 3' (SEQ ID NO:44); and  
5' - CGG CAA TGT TTT ACT GTA GCG CC - 3' (SEQ ID NO:45).

24. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the AmpC beta-lactamase enzyme found in *Citrobacter freundii*.

25. The method of claim 24 wherein the primers are selected from the group of:

5' - CTG GCA ACC ACA ATG GAC TCC G - 3' (SEQ ID NO:18); and  
5' - GCC AGT TCA GCA TCT CCC AGC C - 3' (SEQ ID NO:19).

26. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the AmpC beta-lactamase enzyme found in *Serratia marcescens*.

27. The method of claim 26 wherein the primers are selected from the group of:

5' - CGT GAC CAA CAA CGC CCA GC - 3' (SEQ ID NO:20); and  
5' - CCA GAT AGC GAA TCA GAT CGC - 3' (SEQ ID NO:21).

28. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the plasmid-mediated AmpC beta-lactamase enzymes designated as FOX-1, FOX-2, or MOX-1.

29. The method of claim 28 wherein the primers are selected from the group of:

5' - CCA GCC GAT GCT CAA GGA G - 3' (SEQ ID NO:22); and

5' - CAC GAA CGC CAC ATA GGC G - 3' (SEQ ID NO:23).

30. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the AmpC beta-lactamase enzyme found in *Pseudomonas aeruginosa*.

31. The method of claim 30 wherein the primers are selected from the group of:

5' - GGC ATT GGG ATA GTT GCG GTT G - 3' (SEQ ID NO:24); and

5' - TTA CTA CAA GGT CGG CGA CAT GAC C - 3' (SEQ ID NO:25).

32. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the AmpC beta-lactamase enzyme found in *E. coli*.

33. The method of claim 32 wherein the primers are selected from the group of:

5' - GGA TCA CAC TAT TAC ATC TCG C - 3' (SEQ ID NO:26); and

5' - CGT ATG GTT GAG TTT GAG TGG C - 3' (SEQ ID NO:27).

34. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the K1 beta-lactamase enzyme.

35. The method of claim 34 wherein the primers are selected from the group of:

5' - GCG ACC TGG TTA ACT ACA ATC CC - 3' (SEQ ID NO:28); and

5' - CGG TAG TAT TGC CC TTA AGC C - 3' (SEQ ID NO:29).

36. The method of claim 34 wherein the primers are selected from the group of:

5' - CGG AAA AGC ACG TCG ATG GG - 3' (SEQ ID NO:30); and

5' - GCG ATA TCG TTG GTG GTG CC - 3' (SEQ ID NO:31).

37. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the PSE1, PSE4, and CARB3 beta-lactamase enzymes.

38. The method of claim 37 wherein the primers are selected from the group of:

5' - CTC GAT GAT GCG TGC TTC GC - 3' (SEQ ID NO:32); and

5' - GCG ACT GTG ATG TAT AAA CG - 3' (SEQ ID NO:33).

39. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the OXA-9 beta-lactamase enzyme.

40. The method of claim 39 wherein the primers are selected from the group of:

5' - CGT CGC TCA CCA TAT CTC CC - 3' (SEQ ID NO:34); and

5' - CCT CTC GTG CTT TAG ACC CG - 3' (SEQ ID NO:35).

41. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the OXA-12 beta-lactamase enzyme.

42. The method of claim 41 wherein the primers are selected from the group of:

5' - CGC TGG GAA ACC TAT TCG G - 3' (SEQ ID NO:36); and

5' - CTG CCA TCC AGT TTC TTC GGG - 3' (SEQ ID NO:37).



43. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the OXA-5, 6, 7, 10, 11, 13, and 14 beta-lactamase enzymes.

44. The method of claim 43 wherein the primers are selected from the group of:

5' - GGT GGC ATT GAC AAA TTC TGG - 3' (SEQ ID NO:38); and

5' - CCC ACC ATG CGA CAC CAG - 3' (SEQ ID NO:39).

45. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the OXA-1 beta-lactamase enzyme.

46. The method of claim 45 wherein the primers are selected from the group of:

5' - TGT GCA ACG CAA ATG GCA C - 3' (SEQ ID NO:40); and

5' - CGA CCC CAA GTT TCC TGT AAG TG - 3' (SEQ ID NO:41).

47. The method of claim 17 wherein the primers are specific for nucleic acid characteristic of the OXA-2, 3, and 15 beta-lactamase enzymes.

48. The method of claim 47 wherein the primers are selected from the group of:

5' - AGG CAC GAT AGT TGT GGC AGA C - 3' (SEQ ID NO:42); and

5' - CAC TCA ACC CAT CCT ACC CAC C - 3' (SEQ ID NO:43).

49. A diagnostic kit for detecting a TEM family beta-lactamase which comprises packaging, containing, separately packaged:

(a) at least one primer pair capable of hybridizing to beta-lactamase nucleic acid of interest;

(b) a positive and negative control; and

(c) a protocol for identification of the beta-lactamase nucleic acid of interest.

50. The diagnostic kit of claim 49 wherein the primers are selected from the group of:

5' - TGC TTA ATC AGT GAG GCA CC - 3' (SEQ ID NO:1);

5' - AGA TCA GTT GGG TGC ACG AG - 3' (SEQ ID NO:2);

5' - CTT GGT CTG ACA GTT ACC - 3' (SEQ ID NO:3);

5' - TGT CGC CCT TAT TCC - 3' (SEQ ID NO:4); and

5' - TCG GGG AAA TGT GCG - 3' (SEQ ID NO:5).

51. A diagnostic kit for detecting a SHV family beta-lactamase which comprises packaging, containing, separately packaged:

(a) at least one primer pair capable of hybridizing to beta-lactamase nucleic acid of interest wherein at least one of the primers is selected from the primers of claim 2;

5 (b) a positive and negative control; and

(c) a protocol for identification of the beta-lactamase nucleic acid of interest.

52. A diagnostic kit for detecting an AmpC family beta-lactamase which comprises packaging, containing, separately packaged:

10 (a) at least one primer pair capable of hybridizing to beta-lactamase nucleic acid of interest;

(b) a positive and negative control; and

(c) a protocol for identification of the beta-lactamase nucleic acid of interest.

15 53. The kit of claim 52 wherein at least one of the primers is selected from the group consisting of:

5' - CTG GCA ACC ACA ATG GAC TCC G - 3' (SEQ ID NO:18);

5' - GCC AGT TCA GCA TCT CCC AGC C - 3' (SEQ ID NO:19);

5' - CGT GAC CAA CAA CGC CCA GC - 3' (SEQ ID NO:20);

20 5' - CCA GAT AGC GAA TCA GAT CGC - 3' (SEQ ID NO:21);

5' - GGC ATT GGG ATA GTT GCG GTT G - 3' (SEQ ID NO:24);

5' - TTA CTA CAA GGT CGG CGA CAT GAC C - 3' (SEQ ID NO:25);

5' - GGA TCA CAC TAT TAC ATC TCG C - 3' (SEQ ID NO:26);

5' - CGT ATG GTT GAG TTT GAG TGG C - 3' (SEQ ID NO:27); and  
complements thereof.

5 54. A diagnostic kit for detecting a K1 family beta-lactamase which comprises  
packaging, containing, separately packaged:

(a) at least one primer pair capable of hybridizing to beta-lactamase nucleic acid  
of interest;

(b) a positive and negative control; and

10 (c) a protocol for identification of the beta-lactamase nucleic acid of interest.

55. The kit of claim 54 wherein at least one of the primers is selected from the  
group consisting of:

5' - GCG ACC TGG TTA ACT ACA ATC CC - 3' (SEQ ID NO:28);

15 5' - CGG TAG TAT TGC CC TTA AGC C - 3' (SEQ ID NO:29);

5' - CGG AAA AGC ACG TCG ATG GG - 3' (SEQ ID NO:30);

5' - GCG ATA TCG TTG GTG GTG CC - 3' (SEQ ID NO:31); and  
complements thereof.

20 56. A diagnostic kit for detecting a PSE1, PSE4, or CARB3 family beta-lactamase  
which comprises packaging, containing, separately packaged:

(a) at least one primer pair capable of hybridizing to beta-lactamase nucleic acid

of interest;

(b) a positive and negative control; and

(c) a protocol for identification of the beta-lactamase nucleic acid of interest.

5      57.      The kit of claim 56 wherein at least one of the primers is selected from the group consisting of:

5' - CTC GAT GAT GCG TGC TTC GC - 3' (SEQ ID NO:32);

5' - GCG ACT GTG ATG TAT AAA CG - 3' (SEQ ID NO:33); and

complements thereof.